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TRANSLATION NO. 2/64

DATE: 20 March 1968

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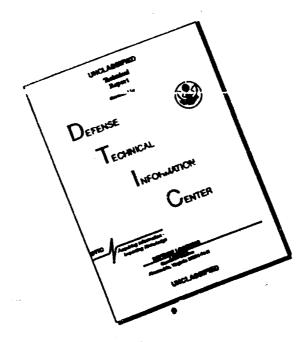
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## NEW CLINICALLY SIGNIFICANT METHOD OF DETERMINING GLYCOPROTEINS IN BLOOD SERUM

Vrachebnoye Delo (Medical Affairs) No. 6, June 1965, Pages 65-69 Ya. N. Dotsenko
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In 1961, S. Ya. Shteynberg and Ya. N. Dotsenko proposed a simple method, requiring no reagents in short supply, for the determination of glycoproteins in the blood serum by means of the reaction with ammonium molybdate. This method of determining glycoproteins consists of the hydrolysis, with trichloacetic acid, of glycoproteins to monosaccharides and proteins, followed by a quantitative determination of the sugars by the colorimetric reaction with ammonium molybdate. Color intensity is determined with a photoelectrocolorimeter, and results of the reaction are expressed in units of optical density. A detailed description of the procedure is given in the periodical Vrachebnoye delo [Medical Practice] (1962, No 12, pages 43-45).

For a comparison of the degree of sensitivity of the ammonium molybdate reaction with that of the widely utilized reaction with diphenylamine, concurrent determinations of glycoproteins were made by both methods in the blood serum of 30 patients of different sex and age and with different disorders. In all the cases, without exception, on study of the same blood sample by these methods the optical density value was found to be higher on determination by the reaction with ammonium molybdate (by 2.5 times on the average). The ratio of optical density values, however, was not constant and varied within wide limits - from 1.66 to 3.28. This is explained by the fact that the reaction with ammonium molybdate is not fully identical with the Dichet reaction. According to published data the diphenylamine reagent reacts only with one of the components contained in the glycoproteins, namely with sialic acid (Hess). Ammonium molybdate apparently reacts with many of the sugars contained in the glycoproteins, reacting differently with each of them.

In 21 out of the 30 patients under study the Dichet reaction values

varied within the normal range (from 0.155 to 0.200), while in the other nine patients the values were found to be moderately elevated (ranging from 0.206 to 0.266). In these same patients the animonium molybdate reaction values were found to be within the normal range only in five of the studies. In 13 patients they were moderately elevated (ranging from 0.416 to 0.500), while in the other 12 patients they were significantly elevated (ranging from 0.504 to 0.638).

The new method for the determination of glycoproteins in the blood serum by reaction with ammonium molybdate is widely utilized, since February 1961, at the faculty's therapeutics clinic of the medical faculty of Khar'kov Medical Institute. In the present communication are analyzed the results of a study of 278 patients with different disorders (rheumatism, hypertension and atherosclerosis, cholecystohepatitis, peptic ulcer, malignant neoformations).

To determine the magnitude of the normal values of the ammonium molybdate reaction we have made a study of 12 healthy students. In them the reaction value ranged from 0.300 to 0.380 (M = 0.332;  $\sigma = \pm 0.032$ ), which we have adopted as the norm.

We undertook the task of ascertaining the diagnostic value of the reaction with ammonium molybdate for determining the activity of the rheumatic process. Values of the reaction with ammonium molybdate were found to be high in all patients -- ranging from 0.505 to 1.000 (M = 0.718;  $\sigma = \pm 0.145$ ).

In 75 patients with rheumatic lesions of the heart, was present a rheumatic process that followed a latent course.

The ammonium molybdate reaction values were found in 12 patients to be within the normal range -- from 0.280 to 0.370 (M = 0.337;  $O \pm 0.035$ ). In 12 other patients these values were slightly elevated, ranging from 0.390 to 0.425 (M = 0.410;  $O \pm 0.011$ ); while in the remaining patients (51 persons) they were elevated, ranging from 0.440 to 0.700 (M + 0.532;  $O \pm 0.071$ ).

Those patients in whom the ammonium molybdate reaction values were found to be within the normal range or slightly elevated (24 persons) had been undergoing, prior to the study, an antirheumatic treatment for a long time (3-4 weeks and longer). We attribute to this fact the decrease of the ammonium molybdate reaction values, as compared with the other patients affected with rheumocarditis following a sluggish course.

In seven patients with rheumatism in the nonactive phase, associated with heart lesion, the ammonium molybdate reaction values were within the normal range -- from 0.270 to 0.350 (M = 0.327;  $S = \pm 0.036$ ).

Dynamic observations of the patients with an active rheumatic process have shown that as the condition of the patients undergoes improvement, the ammonium molybdate reaction values decrease, reverting to the normal level substantially later than the ESR.

As was stated above, changes in the level of blood glycoproteins are not a specific reaction for any particular disorder, but are met with in many disorders. According to published data, the level of blood glycoproteins rises not only in cases of rheumatism, but also in rheumatoid polyarthritis, tuberculosis, pneumonia, radiation sickness, neoformations, hepatites, colitis, atherosclerosis and other disorders.

We made a study of 39 patients with cholecystites and cholecysto-hepatites. The persons under study, mostly young people, were admitted for treatment in connection with exacerbation of pain in the right hypochondrium; also noted were pain in the region of the gallbladder, enlarged and tender liver (in 30 patients), subfeorile body temperature (12 patients), accelerated ESR (in 18 patients). In many of the patients were found disturbances of the liver function: a shift of the coagulation band to the right, in Weltmann's test -- in 21 patients out of 22; positive Takata-Ara reaction -- in 15 patients out of 23; rise of blood bilirubin level -- in 10 patients out of 12; and positive Quick's test -- in 9 patients out of 13.

Ammonium molybdate reaction values were found to be elevated in 34 out of the 39 patients with cholecystitis and cholecystohepatitis (M = 0.506; S = ± 0.067). A parallelism could be noted between the degree of manifestation of inflammatory process exacerbation and the glycoprotein level in the blood semim. The results of our studies are in agreement with the published data (Z. A. Bondar' and I. S. Melkumova, 1960; N. A. Zaslavskaya, 1961; and others).

A study was made of 15 patients with cancer with various localization of primary focus (cancer of the stomach -- in six patients; cancer of the lungs -- in six; and others had cancer of the pancreas, prostate or of the liver). Metastases were present in eight of the patients. In all of the patients, without exception, the ammonium molybdate reaction values were elevated, ranging from 0.440 to 0.980 (M = 0.650; G = + 0.174).

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We divided the patients with peptic ulcer (25 persons) into two groups. The first group included 13 patients with duodenal ulcer in an exacerbation phase, without complications or concurrent disorders. In four of them the ammonium molybdate reaction values were slightly elevated (ranging from 0.400 to 0.416), while in the others they were within the norm  $(M = 0.374; O = \pm 0.038)$ .

The second group consisted of 12 patients with duodenal vicer, in whom a concurrent disorder was observed (chronic cholecystitis or cholecystohepatitis in 11 patients, and marked periduodenitis in one female patient). In two patients of this group the ammonium molybdate reaction values were within the norm (0.315 and 0.350); in the others they were elevated (ranging from 0.400 to 0.590). The difference between the values of these two groups of peptic ulcer patients is not reliable statistically  $(P \le 0.05)$ .

Ammonium molybdate reaction values were found to be elevated (ranging

from 0.400 to 0.800) in 31 out of the 43 patients with atherosclerosis which we have studied. The highest level of blood glycoproteins was found in patients with myocardial infarction, and in patients with cerebral vascular crises, in a pre-infarctal state (M = 0.500;  $G = \pm 0.092$ ); in the other patients of this group M = 0.424;  $G = \pm 0.086$ .

Among the 57 patients with hypertensive vascular disease the normal level of blood glycoproteins was found in 9 patients; a slightly elevated (from 0.400 to 0.400) -- in 24 patients. In the other 24 patients it ranged from 0.500 to 0.800. The highest ammonium molybdate reaction values were found in 11 patients with hypertensive vascular disease who were admitted for treatment with signs of hypertensive crisis (M = 0.516; O = + 0.064), with suspected myocardial infarction and with myocardial infarction (M = 0.573; O = + 0.115) in 11 patients. The level of glycoproteins was appreciably lower in 18 patients admitted for treatment because of increasing headaches or a moderate increase in frequency of the attacks of stenocardia (M = 0.399; O = + 0.044; P < 0.02).

In 29 patients with atherosclerosis and hypertensive vascular disease the blood glycoproteins were studied in their dynamics, two to three times during the period of treatment at the department. In 11 out of the 29 patients the repeat-study showed that ammonium molybdate reaction values had decreased % - 50% (M = 19%) in comparison with the initial level. In all these patients the decrease of blood glycoprotein level coincided with a clinically improved condition. In five of the patients the blood glycoprotein level showed on repeat-study an increase by an increase of the patients arise of blood glycoprotein level coincided with a worsening of their condition. In the other 13 patients of this 29-patient group repeat study showed that the ammonium molybdate reaction values remained unchanged, while a clinical improvement of their condition was observed.

### Conclusions

- 1. The reaction with ammonium molyblate is a simple and at the same time a very sensitive method for determining the level of glycoproteins in the blood serum, which has a number of advantages over the diphenylamine reaction.
- 2. The reaction with ammonium molybdate is a valuable supplementary test for determining the activity of the rheumatic process, and also a checkup test for the assessment of the results of the conducted treatment of patients with rheumatism.
- 3. The utilization of the ammonium molybdate reaction might be found of value in the examination and treatment of patients with disorders of abdominal-cavity organs, in the instances of a differential diagnosis between peptic ulcer and neoformations, as well as between peptic ulcer and cholecystohepatitis.

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